AMENDMENTS TO THE SPECIFICATION:

Please replace the paragraph at p. 9, l. 27 to p. 10, l. 5 with the following amended paragraph:

Moreover, such aforementioned recombinant tissue protective cytokines may be further modified by having a chemical modification of one or more amino acids, such as described in the following co-pending applications: PCT application serial no. PCT/US01/49479, filed December 28, 2001, U.S. Patent Application Serial No. 09/753,132 filed December 29, 2000, and U.S. Patent Application ~~Attorney Docket No. KW00-009C02-US~~ 10/188,905 filed July 3, 2002, each of these applications is incorporated herein by reference in their entirety. These further chemical modifications may be used to enhance the tissue protective activities of the recombinant tissue protective cytokines or suppress any effects the recombinant tissue protective cytokines may have on bone marrow. In a further embodiment, the additional chemical modification is provided to restore solubility of the molecule that may be reduced as a result of the aforementioned genetic modification, such as chemically adding a positive or negative charge to the molecule if a charged amino acid residue is changed to an uncharged residue.

Please replace the paragraph at p. 10, ll. 7-16, with the following amended paragraph:

By way of non-limiting examples, recombinant tissue protective cytokines of the invention include human erythropoietin mutein S100E ~~(SEQ ID NO:5)~~ (SEQ ID NO:62), human erythropoietin mutein K45D ~~(SEQ ID NO:6)~~ (SEQ ID NO:44), and any of the nonerythropoietic yet cellular protective recombinant tissue protective cytokines or those able to benefit a responsive cell, tissue or organ, that are described in Elliott *et al.*, 1997, Blood 89:493-502; Boissel *et al.*, Journal of Biological Chemistry, vol. 268, No. 21, pp. 15983-15993 (1993); Wen *et al.*, Journal of Biological Chemistry, vol. 269, No. 36, pp. 22839-22846 (1994); and Syed *et al.*, Nature, vol. 395, pp. 511-516 (1998), which are incorporated herein by reference in their entireties. The present invention is directed to methods for the use of any of the aforementioned recombinant tissue protective cytokines for the protection, restoration, and enhancement of responsive cells, tissues, and organs.

Please replace the paragraph at p. 16, ll. 9-16, with the following amended paragraph.

According to one aspect of the invention, there is provided an isolated nucleic acid molecule that comprises a nucleotide sequence which encodes a polypeptide comprising the recombinant tissue protective cytokine as described herein above. In one embodiment, the isolated nucleic acid molecule comprises the nucleotide sequence of nucleotide residues 5461 through 6041 of the ~~vecotor contruct~~ vector construct of SEQ ID NO: 208, nucleotide residues 5461 through 6041 of SEQ ID NO: 209, nucleotide residues 5461 through 6041 of SEQ ID NO: 210, nucleotide residues 5461 through 6041 of SEQ ID NO: 211, or nucleotide residues 5461 through 6041 of ~~SEQ ID NO: 212~~ SEQ ID NO:5.

Please replace the paragraph at p. 20, ll. 3-17 with the following amended paragraph:

The invention further provides for the use of a recombinant tissue protective cytokine as described herein above, that lacks at least one erythropoietic activity selected from the group consisting of increasing hematocrit, vasoactive action (vasoconstriction/ vasodilatation), hyperactivating platelets, pro-coagulant activity and increasing production of thrombocytes, for the preparation of a pharmaceutical composition for the protection against and prevention of a tissue injury as well as the restoration of and rejuvenation of tissue and tissue function in a mammal. In one embodiment, the injury is caused by a seizure disorder, multiple sclerosis, stroke, hypotension, cardiac arrest, ischemia, myocardial infarction, inflammation, age-related loss of cognitive function, radiation damage, cerebral palsy, neurodegenerative disease, Alzheimer's disease, Parkinson's disease, Leigh's disease, AIDS dementia, memory loss, amyotrophic lateral sclerosis, alcoholism, mood disorder, anxiety disorder, attention deficit disorder, hyperactivity, autism, ~~Creutzfeld-Jakob~~ Creutzfeldt-Jakob disease, brain or spinal cord trauma or ischemia, heart-lung bypass, chronic heart failure, macular degeneration, diabetic neuropathy, diabetic retinopathy, glaucoma, retinal ischemia, or retinal trauma.

Please replace the paragraph at p. 39, l. 8 to p. 40, l. 14 with the following amended paragraph:

Recombinant tissue protective cytokines of the invention include erythropoietin muteins, that maintain partial or full erythropoietic activity. Erythropoietin is a glycoprotein

hormone which in humans has a molecular weight of about 34 kDa. The mature protein comprises 165 amino acids (SEQ ID NO:10), and the glycosyl residues comprise about 40% of the weight of the molecule. The forms of recombinant tissue protective cytokine useful in the practice of the present invention encompass at least a single amino acid change in naturally-occurring, synthetic and recombinant forms of the following human and other mammalian erythropoietin-related molecules: erythropoietin, asialoerythropoietin, deglycosylated erythropoietin, erythropoietin analogs, erythropoietin mimetics, erythropoietin fragments, hybrid erythropoietin molecules, erythropoietin receptor-binding molecules, erythropoietin agonists, renal erythropoietin, brain erythropoietin, oligomers and multimers thereof, and congeners thereof. Such equivalent recombinant tissue protective cytokines include mutant erythropoietins, which may contain substitutions, deletions, including internal deletions, additions, including additions yielding fusion proteins, or conservative substitutions of amino acid residues within and/or adjacent to the amino acid sequence, but that result in a "silent" change, in that the change produces a functionally equivalent erythropoietin mutein or recombinant tissue protective cytokine. In a preferred embodiment, the recombinant tissue protective cytokine is nonerythropoietic, i.e. lacking or exhibiting diminished erythropoietic activity. Conservative amino acid substitutions may be made on the basis of similarity in polarity, charge, solubility, hydrophobicity, hydrophilicity, and/or the amphipathic nature of the residues involved. For example, nonpolar (hydrophobic) amino acids include alanine, leucine, isoleucine, valine, proline, phenylalanine, tryptophan, and methionine; polar neutral amino acids include glycine, serine, threonine, cysteine, tyrosine, asparagine, and glutamine; positively charged (basic) amino acids include arginine, lysine, and histidine; and negatively charged (acidic) amino acids include aspartic acid and glutamic acid. Alternatively, non-conservative amino acid changes, and larger insertions and deletions may be used to create functionally altered recombinant tissue protective cytokines. Such mutants can be used to alter erythropoietin properties in desirable ways. For example, in one embodiment, an erythropoietin useful for the practice of the invention can be a recombinant tissue protective cytokine altered in one or more amino acids within the four functional domains of erythropoietin which affect receptor binding: VLQRY (SEQ ID NO:1) and/or TKVNFYAW (SEQ ID NO:2) and/or SGLRSLTTL (SEQ ID NO:3) and/or SNFLRG (SEQ ID NO:4). In another embodiment, erythropoietins containing mutations in the surrounding areas of the molecule which affect the kinetics or

receptor-binding properties of the molecule can be used. Determining which alterations, or which positions in the domains will effect binding can be accomplished using standard methods. For example, the domains may be altered by pair-wise alanine mutations (ala-scanning mutagenesis) followed by measurement of binding kinetics of mutants to examine the effect on binding to a receptor (Bernat *et al.*, 2003, PNAS 100:952-957; Wells *et al.*, 1989, Science 244:1081-1085).

Please replace the paragraph at p. 42, ll. 1-23 with the following amended paragraph:

Furthermore, derivative recombinant tissue protective cytokine molecules desirable for the uses described herein may be generated by guanidination, amidination, carbamylation (carbamoylation), trinitrophenylation, acylation such as acetylation or succinylation, nitration, or modification of arginine, aspartic acid, glutamic acid, lysine, tyrosine, tryptophan, or cysteine residues or carboxyl groups, among other procedures, such as limited proteolysis, removal of amino groups, and/or mutational substitution of arginine, lysine, tyrosine, tryptophan, or cysteine residues by molecular biological techniques to produce erythropoietin muteins or recombinant tissue protective cytokines which maintain an adequate level of activities for specific organs and tissues but not for others, such as erythrocytes (*e.g.*, Satake *et al*; 1990, *Biochim. Biophys. Acta* 1038:125-9; incorporated herein by reference in its entirety[[)]], in which *in vivo* biological activity was determined by the polycythemic mouse bioassay). One non-limiting example as described hereinbelow is the modification of erythropoietin arginine residues by reaction with a glyoxal such as phenylglyoxal (according to the protocol of Takahashi, 1977, *J. Biochem.* 81:395-402). As will be seen below, such a recombinant tissue protective cytokine molecule fully retains the neurotrophic effect of erythropoietin. Such recombinant tissue protective cytokine molecules are fully embraced for the various uses and compositions described herein. In addition, these chemical modifications may be further used to enhance the protective effects of the recombinant tissue protective cytokines or neutralize any changes in the charge of the molecule resulting from the amino acid mutation of the native erythropoietin. Such modifications are described in co-pending applications:[[,]] serial no. PCT/US01/49479, filed December 28, 2001; serial no. 09/753,132, filed December 29, 2000 and ~~Attorney's Docket~~

~~No. KW00-009C02-US~~ serial no. 10/188,905, filed July 3, 2002, all of which are incorporated herein in their entireties.

Please replace the paragraph at p. 47, ll. 11-22 with the following amended paragraph:

Further to the above-mentioned erythropoietin modifications useful herein, the following discussion expands on the various recombinant tissue protective cytokines of the invention. As described in Elliott *et al.*, Boissel *et al.*, and Wen *et al.*, mentioned above, the following erythropoietin muteins are useful for the purposes described herein, and may be provided in a pharmaceutical composition for the methods herein. In the mutein nomenclature used throughout herein, the changed amino acid is depicted with the native amino acid's one-letter code first, followed by its position in the erythropoietin molecule, followed by the replacement amino acid one-letter code. For example, "human erythropoietin S100E" or "recombinant ~~tisue protectiv~~ tissue protective cytokine S100E" refers to a human erythropoietin molecule in which, at amino acid position 100 of the mature erythropoietin, a serine has been changed to glutamic acid. Such muteins useful for the practice of the present invention include but are not limited to human erythropoietin with at least one of the following amino acid changes:

Please replace the paragraph at p. 68, ll. 6-25 with the following amended paragraph:

Various neuropsychologic disorders which are believed to originate from excitable tissue damage are treatable by the instant methods. Chronic disorders in which neuronal damage is involved and for which treatment by the present invention is provided include disorders relating to the central nervous system and/or peripheral nervous system including age-related loss of cognitive function and senile dementia, chronic seizure disorders, Alzheimer's disease, Parkinson's disease, dementia, memory loss, amyotrophic lateral sclerosis, multiple sclerosis, tuberous sclerosis, Wilson's Disease cerebral and progressive *supra*nuclear palsy, Guam disease, Lewy body dementia, prion diseases, such as spongiform encephalopathies, *e.g.,* Creutzfeldt-Jakob disease, Huntington's disease, myotonic dystrophy, ~~Freidrich's~~ Friedrich's ataxia and other ataxias, as well as Gilles de la Tourette's syndrome, seizure disorders such as epilepsy and chronic seizure disorder, stroke, brain or spinal cord trauma, AIDS dementia, alcoholism, autism, retinal ischemia, glaucoma, autonomic function

disorders such as hypertension and sleep disorders, and neuropsychiatric disorders that include, but are not limited to, schizophrenia, schizoaffective disorder, attention deficit disorder hyperactivity, dysthymic disorder, major depressive disorder, mania, obsessive-compulsive disorder, psychoactive substance use disorders, anxiety, panic disorder, as well as unipolar and bipolar affective disorders. Additional neuropsychiatric and neurodegenerative disorders include, for example, those listed in the American Psychiatric Association's Diagnostic and Statistical Manual of Mental Disorders (DSM), the most current version, IV, of which in incorporated herein by reference in its entirety.

Please replace the paragraph at p. 89, ll. 14-20 with the following amended paragraph:

The PCR product was cloned between the Xho I and Xba I sites in pCiNeo mammalian expression vector (Promega). The clones were sequenced and the sequence was verified to match the sequence in NM_000799 with the exception of a single base. Base 418 in the coding sequence (starting the numbering from the ATG) was C instead of G, changing amino acid 140 in the full length EPO sequence (*i.e.*, SEQ ID NO:6) starting from the first methionine from Arg to Gly. This is, however, normal sequence variation from the original sequence and present in most forms of erythropoietin.

Please replace the three paragraphs at p. 90, ll. 4-10 with the following amended three paragraphs:

This cDNA codes for the full length amino acid sequence of erythropoietin, which is below

MGVHECPAWLWLLLSLLSLPLGLPVLGAPPRLICDSRVLERYLLEAKEAENIT TGCAEHCSLNENITVPDTKVNFYAWKRMEVGQQAVEVWQGLALLSEAVLRGQALL VNSSQPWEPLQLHVDKAVSGLRSLTTLLRALGAQKEAISPPDAASAAPLRTITADTFR KLFRVYSNFLRGKLKLYTGEACRTGDR ~~(SEQ ID NO: 10)~~ (SEQ ID NO:6).

The first 27 amino acid residues of ~~SEQ ID NO:10~~ SEQ ID NO:6 comprise a leader sequence.

Please replace the paragraph at p. 101, ll. 19-25 with the following amended paragraph:

The following are examples of constructs that were made: human EPO(hEPO)-6xHisTag-pCiNeo sequence (SEQ ID NO: 208); hEPO6xHisTag-A30N/H32T-pCiNeo (SEQ ID NO: 209); hEPO-6xHisTag-K45D-pCiNeo sequence (SEQ ID NO: 210); hEPO-6xHisTag-S100E-pCiNeo sequence (SEQ ID NO: 211); and hEPO-6xHisTag-K45D/S100E-pCiNeo sequence ~~(SEQ ID NO: 212)~~ (SEQ ID NO:5). The pCI-neo mammalian expression vector carries the human cytomegalovirus (CMV) immediate-early enhancer/promoter region to promote constitutive expression of cloned DNA inserts in mammalian cells.

Please replace the table at pp. 69-71 with the replacement table submitted herewith as Appendix A. The table is amended to replace the term "Guillian Barre" with the corrected term "Guillain Barre" (see specification as filed at p. 71, l. 13).